The Journal of Experimental Biology 207, 4195-4203
Published by The Company of Biologists 2004
doi:10.1242/jeb.01276

Allometry measurements from *in situ* video recordings can determine the size and swimming speeds of juvenile and adult squid *Loligo opalescens* (Cephalopoda: Myopsida)

Louis D. Zeidberg

Monterey Bay Aquarium Research Institute, 7700 Sandholdt Road, Moss Landing, CA 95039-9644, USA

e-mail: zelo@mbari.org

Accepted 6 September 2004

Summary

Juvenile and adult *Loligo opalescens* Berry were video taped in Monterey Bay with the remotely operated vehicle (ROV) *Ventana*, captured with an otter trawl in Santa Monica Bay, California, and adults were taken from the Monterey Bay fishery. Behavioral observations were made over a 13 h period of video sequences. Allometry measurements were made on 157 squids ranging in size from 12 to 151 mm mantle length (*ML*). In addition to *ML* we measured the morphometric characters of fin length (*FL*), fin width (*FW*), mantle width (*MW*), eye diameter (*ED*), head width (*HW*), funnel aperture diameter (*FA*), fourth arm length (*AL*) and tentacle length (*TL*). *Loligo opalescens* changes shape with ontogeny due to negative allometric growth of ED, HW, TL, MW, FA and positive allometric growth of AL, FL and fin area. The allometry measurements were used to determine the size of juvenile squids video-taped in open water. A linear regression can predict dorsal *ML* in mm from a dimensionless ratio of *ML* upon *ED* (r^2=0.857, P<0.001). Sizes and velocities of video-taped animals were estimated from 26 video sequences ranging from <1.0 to 8 s. The average velocity for squids ranging from 12–116 mm *ML* was 0.21 m s^{-1} and the maximum velocity was 1.60 m s^{-1} (116 mm *ML*). Allometric measurements can provide scale for 2-dimensional images in order to estimate size, velocity and age of animals.

Supplementary material available online at
http://jeb.biologists.org/cgi/content/full/207/24/4195/DC1

Key words: squid, *Loligo opalescens*, allometry, juveniles, *in situ* video, behavior, ecology.

Introduction

Market squid is the largest fishery in California, worth up to $36 million and 118,000 tons annually; it is imperative that the entire life cycle of the species is understood. Previous fieldwork on *Loligo opalescens* has focused on sub-samples of fishery-captured specimens. Two recent exceptions have investigated the behavior and ecology of paralarvae (Zeidberg and Hamner, 2002) and the effects of the El Niño on growth (Jackson and Domeier, 2003). The juvenile phase of *Loligo opalescens* has been described only once previously, when less than 1 kg of juveniles were captured in one of 1375 midwater trawls in the California Current (Mais, 1974).

The juvenile life stages are the least studied phase of most cephalopods. Most of the literature describing juvenile cephalopods, especially the teuthids, comes from laboratory studies (Yang et al., 1986; Hatfield et al., 2001). This is because the juvenile age group is difficult to capture and harder still to observe in the wild. Juveniles are nearly as fast as adults but are not distracted by reproductive activities. Wormuth and Roper (1983) demonstrate that the predicted biomass of squid from net studies far underestimate similar predictions based upon stomach content studies of squid predators. Mais (1974) used large midwater trawls towed at a speed of 3 knots to sample pelagic species in the California Current. He caught adult squid in 36% of 1375 tows, but usually only 1–20 squid were captured per tow, despite towing through schools of squid up to 400 m in diameter. *Loligo opalescens* is obviously adept at avoiding midwater trawl nets, when compared to captures by purse seines, where individual boats can haul >100 tons in one night (L. D. Zeidberg, W. M. Hamner, N. Nezlin and A. Henry, manuscript submitted). Mais (1974) had even greater difficulty netting juveniles than adults. Teuthids are also quite difficult to rear and consequently only a few laboratory experiments have been conducted that included juvenile squid (Yang et al., 1986; Hatfield et al., 2001). Boletsky (1987) reviewed the behavior of juvenile cephalopods and noted that 'there are virtually no field studies on the behavior of very young cephalopods', and the state of the literature has changed little in 17 years.

Laboratory studies have provided a large amount of information about squid swimming. Packard (1969) first described jetting behavior in *Loligo vulgaris* ranging from 6–320 mm mantle length (*ML*). O'Dor (1982, 1988a) measured

swimming and oxygen metabolism of *Loligo opalescens* in a respirometer and then calculated the forces acting upon the swimming squid. The laboratory study of squid locomotion has advanced recently, including analysis of the jetting capabilities of developing *Sepioteuthis lessoniana* (Thompson and Kier, 2002), thrust from jets, fins and angle of attack of *Lolliguncula brevis* (Bartol et al., 2001), and a description of unsteady motion during jetting and mantle refilling in *Loligo pealei* (Anderson and Demont, 2000).

Studies from field-collected squid demonstrate the importance of understanding the caveats of studying laboratory-reared squid. Jackson and Domeier (2003) analyzed the growth rates and age of *Loligo opalescens* captured in southern California from measurements of *ML*, weight and statolith ring counts. They found a positive correlation between *ML* and colder water and between *ML* and the upwelling index. This contrasts with all of the laboratory results where squid grow faster and larger in warmer water with constant food resources (Hatfield et al., 2001), most likely because squid prey is more abundant in the eastern Pacific Ocean in colder upwelled water. There have been no *in situ* quantitative analyses of teuthid swimming. Although squid have been observed in the open ocean from remotely operated vehicles (ROVs; Hunt et al., 2000; Hunt and Seibel, 2000), this technology has only recently been developed, and there is not yet a method of quantifying size of animals in the open ocean from 2-dimensional video recordings. Here I use allometry measurements in conjunction with video recordings obtained from a ROV to quantitatively investigate *in situ* swimming of *Loligo opalescens*.

Allometry is the study of change in the proportion of various body parts of an organism as a result of growth, and allometric measurements of squid have been examined several times. Zuev (1964) first remarked that squid paralarvae have a bell-shaped mantle, globular fins, large eyes and short arms, whereas adult squid have a long slender mantle, triangular or rhomboid fins, eyes that are smaller in proportion to total length and longer arms than paralarvae. Okutani (1987) evaluated allometry of net-captured *Gonatus madokai*, noting that fin and arm dimensions had positive allometric growth compared to the mantle length, whereas mantle width, head width, eye diameter and tentacle length had inflection points shifting from negative allometric growth to positive at *ML* 22 mm. Vidal (1994) studied allometric growth in *Illex argentinus* (1–55 mm *ML*) and found changes in growth rates similar to *Gonatus madokai* at 28 mm *ML*. Moltschaniwskyj (1995) found the relative growth of the fins, arms and tentacles to be greater in smaller *Photololigo* sp. Rodhouse and Piatkowski (1995) argue that the positive allometric growth of the brachial crown serves to allow rapidly growing teuthids to capture zooplankton of sizes increasing by orders of magnitude as the squid develops. O'Dor and Hoar (2000) use allometry measurements to argue that geometry does not limit growth in teuthids as it does in fish, and they suggest that sexual maturation probably terminates somatic growth.

Herein for the first time *in situ* behavioral observations of a juvenile squid are reported and allometry measurements are used as a tool to determine the size and swimming speeds of squid in images with no backgrounds for size reference. Allometry of *ML* and eye diameter (*ED*) of *Loligo opalescens* is used to quantify the size and swimming speed of juvenile squid in blue water. Video analysis of animal behavior in marine environments has previously been strictly qualitative in nature, due to an inability to determine the scale of images from 2-D photographs. Use of allometry, however, permits size determination and this technique can have wide applications for video analysis, ecological assessments of biomass and individual identification.

Materials and methods

Juvenile and adult *Loligo opalescens* Berry 1911 squid were captured in an otter trawl within 1 m of the bottom of the sandy shelf of Santa Monica Bay, California, USA on 29 occasions between March 8, 1998 and February 12, 2002 aboard the R/V *SeaWorld UCLA* during educational cruises. All tows were made between 10.00 h and 13.00 h. The mouth opening of the otter trawl was 5 m wide and 1 m deep. The 5 min otter trawls were conducted 3 nautical miles southwest (240°) of Marina del Rey, California over sandy bottom substrate. Dates of capture were recorded and the squid were frozen and returned to the laboratory where morphometric measurements were made. The lengths of the dorsal mantle (*ML*), fin (*FL*), funnel aperture (*FA*), fourth arm (*AL*), tentacle (*TL*), diameter of the eye (*ED*), and widths of the fin (*FW*), mantle (*MW*) and head (*HW*) were measured with an ocular micrometer or a caliper to 0.1 mm or 0.5 mm significant digits, respectively. Dimensions of measurements are depicted in Fig. 1. The dorsal mantle length is from the mantle tip to the end of the gladius superior to the head. Fin length is from the points of insertion at the mantle tip to the other insertion point midway on the mantle. Fin width is from the fin tip perpendicular to the point of insertion into the mantle. Eye diameter is the longest distance along the anterior–posterior axis of the slightly oval-shaped eyes. Mantle diameter is the width of the mantle at the point of greatest girth. Calipers were inserted into the funnel aperture and spread until the funnel sides stretched flat, and aperture diameter was derived from the half-circumference dimension. Arms and tentacles were measured when straightened, taking care not to stretch the elastic tissues. The head width is from the center of one eye to the other. I measured these dimensions for 157 squid that ranged in *ML* from 12 mm to 151 mm.

Regression analysis was used to compare the dimensions of the *Loligo opalescens* squid as they developed from juveniles to adults. The allometric equation is $y=ax^b$, where y is the length of the body dimension of interest, x is the *ML*, b is the allometric constant and a is the initial index. The body dimensions and the *ML* were log-transformed and a and b were then determined by linear regression. The allometric factor, b, was >1 if the given body dimension grew at a rate faster than the *ML*, called positive allometric growth. If b=1 then the two dimensions had isometric growth, and if b<1 then *ML* was

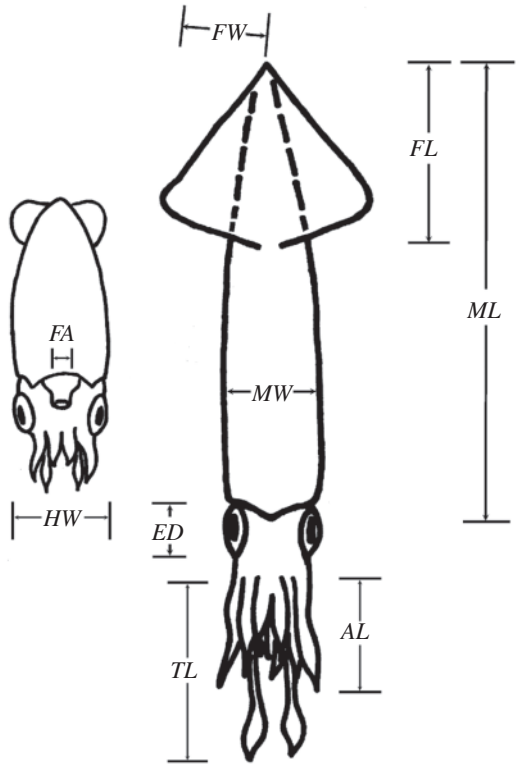

Fig. 1. Dorsal view of *Loligo opalescens* (40 mm *ML*), ventral view of 12 mm *ML* inset to left. Dimensions of each body part measured for allometry analysis: dorsal mantle length (*ML*), fin length (*FL*), funnel aperture (*FA*), fourth arm length (*AL*), tentacle length (TL), diameter of the eye (*ED*), fin width (*FW*), mantle width (*MW*) and head width (*HW*).

growing at a rate faster than the given body dimension – called negative allometric growth.

Juvenile *Loligo opalescens* were video recorded in Monterey Bay, California. High definition video was recorded using a Sony DXC3000 3 chip Camera c/w Fujinon Zoom Lens f1.7 5.5–47 mm mounted aboard the ROV *Ventana* and launched from the R/V *Pt. Lobos* of the Monterey Bay Aquarium Research Institute (MBARI). Global positioning system (GPS), conductivity temperature depth (CTD) and O_2 concentration data were also recorded by the ROV. Video recordings taken at night during the cruises of April 26, August 23, 24 and 25, 2000, were dubbed onto miniDV digital-video tapes. 13.5 h of video tape were analyzed. All of the tape was analyzed for discrete behavioral components while only appropriate sequences were captured (see below) for motion analysis.

Images in which the profile of the squid was clearly visible, the squid jetted parallel to the field of view, and the ROV was hovering, were used to quantify the scale of the animal in the frame. Although there is little distortion from the lens, frames of video images in which the squid was near the edge of the field of view were not analyzed. Twenty-six video sequences were analyzed ranging in duration from 0.66 s to 8 s. Digital video was captured from a Sony 3ccd video camera, DCR-TRV 900, *via* an Orangelink firewire cardbus PC card (OrangeMicro Inc., Anaheim, CA, USA) onto the hard drive of a personal computer where motion analysis was conducted. The video frames were not compressed. IrfanView shareware (version 3.91, Irfan Skiljan, 2700 Wiener Neustadt, Austria) converted the individual frames of the video sequences to bitmap images with a 480-pixel height and 720-pixel width. ImageJ shareware (NIH, http://rsb.info.nih.gov/ij) was then used to quantify the swimming speeds of the individual squids.

Using ImageJ, *ML* and *ED* were measured in pixels by drawing a line over these body parts in 4–16 frames of the video sequences. The mantle length and eye diameter body dimensions were both chosen because of their conspicuousness, their incompressibility, and their similar orientation in the anterior–posterior axis. An average of the measurements of the ratio of mantle length divided by eye diameter (*ML*/*ED*) was used to determine the size of an individual in mm. The regression of dorsal *ML* from the ratio of *ML*/*ED* allowed the estimation of the size of the squid in each sequence. Thus mantle length in pixels was converted to mm and then the total length of the individual was derived from a linear regression of the *ML*.

The ImageJ marking tool was used to give an x and y coordinate to the mantle tip of the squid and a passive particle in the same depth of field as the squid in each frame. The standard distance equation was used to determine the distance in pixels that the passive particle moved between frames, and this distance was subtracted from the distance that the squid had moved to yield a distance corrected to the movements of the ROV. Total length of the squid in pixels was also measured in each frame using ImageJ. The total length in mm (estimated from the allometry regression) was divided by an average of the total length between the frame viewed and the previous frame in pixels, yielding *TL* (mm/pixels). This ratio (mm/pixels), was then multiplied by the corrected distance moved between frames (pixels), yielding an instantaneous distance (mm) for each frame. Velocity was then calculated from the instantaneous equation using 1/29.97 s as the time period between consecutive frames (these video images are recorded at a rate of 29.97 frames s^{-1}).

The x- and y-coordinates in the plane of a video image correspond to the x- and z-axes normally considered in physical oceanography calculations and ignore the oceanographic y-axis, or depth of field. This problem is always encountered when attempting to resolve the movements of animals in three dimensions as captured on 2-dimensional video images. To address this, sequences were chosen in which the animals moved largely parallel to the plane of view, across the screen with minimal change in apparent size. Furthermore, any movement in the direction of the y-axis (toward or away from the camera) would cause an underestimation of the distances that the animals had moved. Thus observations of velocities and swimming capabilities are probably underestimates of *in situ* capabilities.

Three adult squid were video taped by the ROV *Ventana* while on deck of the R/V *Pt. Lobos* in order to confirm the

regression estimates of size and speed. These squid were taken from the Monterey Bay fishery and measured with a caliper for the allometric dimensions. The actual lengths of the squid were compared to estimates of size from video stills. To confirm *in situ* estimates of speed, the squid were attached to a Stanley 25 foot (~7.6 m) tape measure, with rubber bands. The squid were placed upon a 122 cm long table and were video taped 1 m away from the camera housing. One meter of tape measure was paid out and video of the spring-loaded return was recorded. Actual speed was quantified by dividing the distance traveled by the number of frames (1/29.97 s) required for the recoil. Estimates of size and speed from the allometry regression were compared to the measured sizes and speeds.

The ROV *Ventana* camera is calibrated for underwater viewing, but the confirmation measurements were made in air. Normally the optics ratio for water:air is 4:3. That is, a 9 foot long hammerhead shark will look as if it is 12 foot long to a diver when observed underwater. Because all of the measurements made in this investigation are scaled frame by frame and estimated from ratios of two measurements of the animal within each frame, one does not need to account for the water:air increase in perceived size, which is minor compared to the apparent enlargement from a zooming lens.

Results

Behavior, ecology and locomotion

Mantle lengths for squid collected by otter trawl from Santa Monica Bay ranged from 12–147 mm, and for fishery squid ranged from 122–151 mm. Fig. 2 shows the dates of capture and the *ML* of the squid captured in southern California. There is a missing size cohort (55–85 mm *ML*), which is shaded in grey. The greatest landings of the commercial fishery in the Southern California Bight occur from October to May.

Juvenile squid were present 13.4% (132/985 min) of the time in front of the camera during dives that occurred exclusively over the shelf area north of the Monterey Canyon in 0–50 m of water at night. Individual squid spent significantly less time in front of the camera than did shoals (groups of squids in non-parallel orientation), averaging 8.7 and 43.4 s, respectively (Mann–Whitney *U*-test, P=0.001). Shoals averaged 19 individuals but with large variability in size (S.D.=25, N=171). The only significant correlation for the physical parameters measured was with longitude, r^2=0.225, P<0.001, i.e. there were more juvenile squid on camera further away from the eastern shore of the Monterey Bay.

The behavioral repertoire of juvenile *Loligo opalescens* is similar to that of the adults. I observed ten of the 13 chromatic components and all seven of the postural displays previously described (Hunt et al., 2000). Three chromatic components, however, were absent (dark keel, countershading and armstripe), but two new displays were noted (double chevron and white stripe, Fig. 3). The double chevron display consisted of two dark V-shaped patterns on the dorsal surface of the mantle in conjunction with an eye band. The white stripe display extends the entire length of the mantle, surrounded by an otherwise all-dark dorsal mantle.

In addition to discrete postures, I noted two new swimming capabilities. *Loligo opalescens* were observed to initiate movement from hovering in place by bending their arms perpendicular to the anterior–posterior axis and then snapping the arms back to a position parallel with the long axis of the body, in conjunction with a mantle inhalation and then a jet escape [movie 1, c_start; available as supplementary material; video footage ©2000 MBARI].

Toroids, small turbulent packages of water that rolled up into a coherent vortex ring, associated with rapid jetting, were observed on three occasions. In all cases the *Loligo opalescens* were jetting backwards fast, then stopped by braking with their fins and bending their funnel back toward the mantle tip to jet forward [movie 2, Toroid; available as supplementary material; video footage ©2000 MBARI]. Ink was ejected during these maneuvers, allowing *in situ* dye studies of the jet expulsion.

I estimated that the *ML* of *Loligo opalescens* ranged from 12 to 116 mm in the 26 analyzed video sequences. Using my

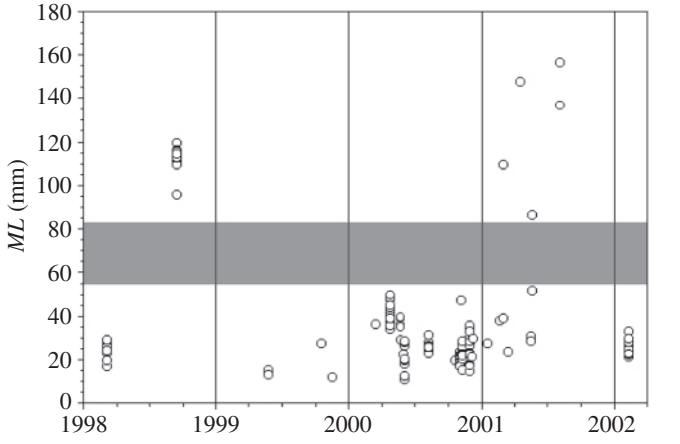

Fig. 2. Date and dorsal mantle length *ML* of each *Loligo opalescens* squid captured with the otter trawl in Santa Monica Bay from March 1998 to February 2002. The missing size range (55–85 mm *ML*) in all 5 years is shaded grey. One to two cohorts exist in each year; fishing for spawning adults occurs near this area from October to May.

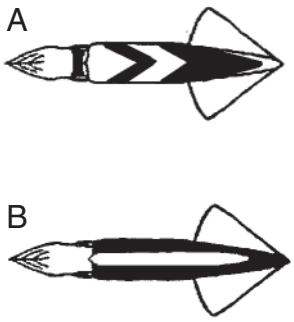

Fig. 3. *Loligo opalescens* chromatic displays observed in juveniles but not in adults. (A) Double chevron; (B) white stripe.

allometry regression (results below) I estimated velocities for squid that were moving by both jet-and-sink and the climb-and-glide swimming gaits (as described in O'Dor, 1988a). One jet-and-sink sequence [Fig. 4; movie 3, jet_sink; available as supplementary material; video footage ©2000 MBARI] illustrates the coordinated timing of vertical position in the field of view, velocity and mantle contraction. For all sequences, maximum and mean velocity (m s^{-1}) scaled well with mantle length (mm), thus V_{max}=0.013*ML*; r^2=0.857, V_{mean}=0.005*ML*; r^2=0.75, calculated from linear regressions where the intercept was forced through zero. The average velocity was 0.21 m s^{-1} (5.15 *ML* s^{-1}) for all video sequences, and the greatest instantaneous velocity was 1.60 m s^{-1} (11.78 *ML* s^{-1}) for the largest squid. The Reynolds number for these squids ranged from 222 to 32 638, using mantle width for *l*, velocity estimated from the regression *u*, and 1.35×10^{-6} for kinematic viscosity υ, in water at 10°C (Denny, 1993).

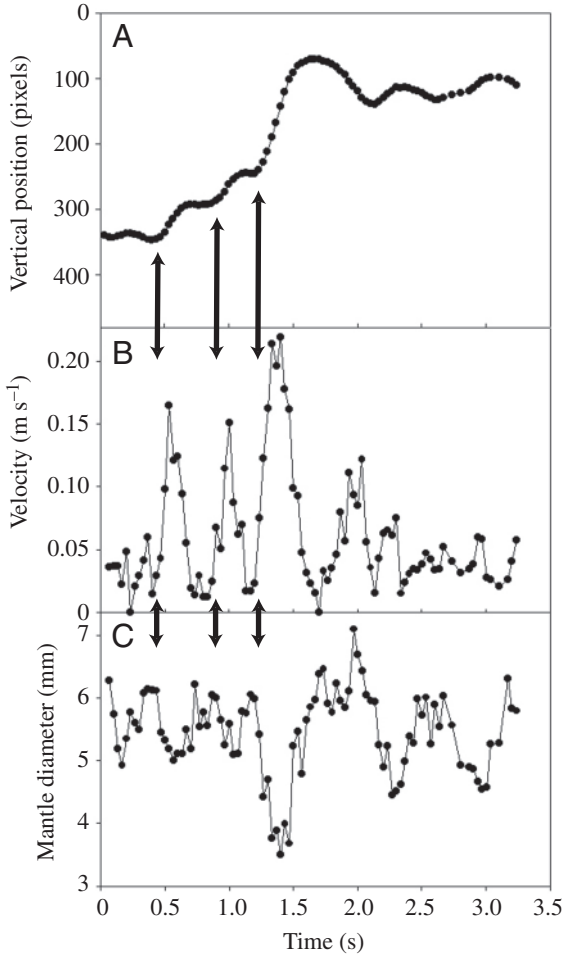

Fig. 4. Jet-and-sink behavior for a squid with an estimated mantle length of 18.55 mm for a 3.5 s video sequence [movie 3, jet_sink; available at supplementary material; video footage ©2000 MBARI]. (A) The vertical position of the mantle tip within the frame (*y* coordinate of pixel). (B) The velocity of the squid in each frame. (C) Mantle diameter (mm). In B and C, pixels are converted to m s^{-1} and mm *via* the allometry regression of Fig. 7A. Increases in velocity and vertical position corresponds to mantle contractions (arrows), with the greatest increase in vertical position and velocity, and the smallest mantle diameter, at time 1.4 s.

Allometry measurements

The results of the body dimension measurements of *Loligo opalescens* from the allometric equation, $y=ax^b$, are summarized in Table 1. The eye diameter (*ED*), head width (*HW*), tentacle length (*TL*), mantle width (*MW*) and funnel aperture diameter (*FA*) have negative allometric growth, growing proportionately slower than *ML*. The arm length (*AL*), fin length (*FL*) and fin area have positive allometric growth. *TL* and *FW* were not significantly different from isometry (*F*-test, *P*>0.05). When comparing an area measurement to a linear measurement, a slope of 2 would demonstrate isometric growth. The surface area of the fins [$2(0.5\times FL\times FW)$] had a positive allometric growth factor of 2.361 compared to *ML* (linear regression, r^2=0.957, *P*<0.001, Fig. 5) – the fins get larger in relation to length with age. The diameter of the funnel aperture had negative allometric growth factor of 0.803 (Fig. 6), thus as squid develop, fin area increases and funnel aperture diameter decreases.

Allometry for scale

Dorsal mantle length (*ML*) can be estimated from a ratio of measured *ML* upon eye diameter (*ED*). Measurements of 157 squid ranging in mantle length from 12–151 mm were used to calculate this linear regression *ML*=–85.285+18.2052(*ML*/*ED*); r^2=0.857, *F*-test *P*<0.001, (Fig. 7A). Because ML/ED (mm/mm) is a dimensionless ratio, the size of a squid in any image where the mantle length and eye diameter are clearly visible can be estimated using this regression by measuring the length of lines, in pixels, that are drawn over these body dimensions. With this regression, the ratio in pixels can then be converted to a dorsal *ML* in mm and thus the size of squid videotaped in blue water can be estimated. Total length *TL* can

Table 1. *Results of allometry analysis for body dimensions of* Loligo opalescens *ranging in* ML *from 12–147 mm*

	a	b	r^2	*P*<0.05
Eye diameter	–0.334	0.673	0.933	*
Head width	–0.030	0.619	0.861	*
Mantle width	0.021	0.655	0.844	*
Funnel diameter	–1.053	0.803	0.758	*
Arm length	–0.728	1.117	0.902	*
Tentacle length	–0.025	0.934	0.797	NS
Fin length	–1.094	1.372	0.968	*
Fin width	–0.870	1.025	0.877	NS
Fin area	–2.214	2.361	0.957	*

The allometric growth equation $y=ax^b$ where *y* is the particular body dimension and *x* is *ML*, log-transformed and analyzed with linear regression. r^2 is the correlation coefficient, a is the intercept (initial index) and b is the slope (allometric growth factor). The allometric growth factor, b, is significant by *F*-test for all body dimensions except fin width and tentacle length. NS, not significant.

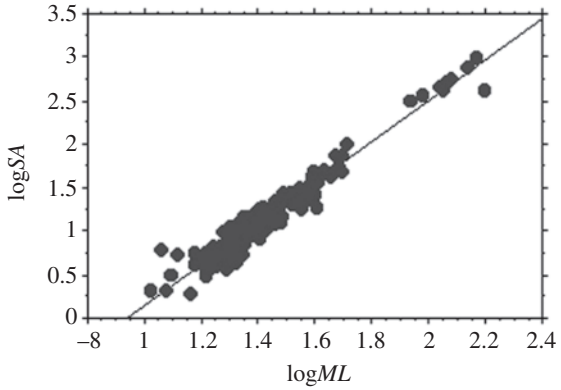

Fig. 5. The fins of *Loligo opalescens* are triangular; surface area (*SA*) of both fins=2(0.5×fin width×fin length at the base). The surface area of both fins provides lift that increases exponentially with the dorsal mantle length *ML* of the developing squid. y=2.361x–2.214; r^2=0.957, P<0.0001, N=136. The slope of the linear regression line is 2.361, demonstrating positive allometry, i.e. slope is significantly different than 2.

be estimated from mantle length. *TL*=–4.0244+1.6227*ML*; r^2=0.989, P<0.001, N=157 (Fig. 7B).

Three squids of known size were attached to a tape measure and videotaped from the ROV *Ventana* while of the deck of the R/V *Pt. Lobos*. There were two males, 126 and 151 mm *ML*, and one female, 124 mm *ML*. The 126 mm *ML* male moved 0.87 m s^{-1} as the tape measure retracted. From the allometry regression I estimated his size as 131 mm *ML* and he was estimated to move 0.89 m s^{-1} (5.7% and 2.2% errors, respectively). The 151 mm *ML* male had a speed of 0.58 m s^{-1} as the tape measure recoiled; the allometry regression predicted his *ML* to be 153 mm and his speed as 0.64 m s^{-1} (1.5% and 9.2% errors, respectively). The allometry regression estimated the female to be 124 mm *ML* and to have moved with a speed of 0.92 m s^{-1} *versus* an actual speed of 0.88 m s^{-1} (0.19% and 4.3% errors, respectively).

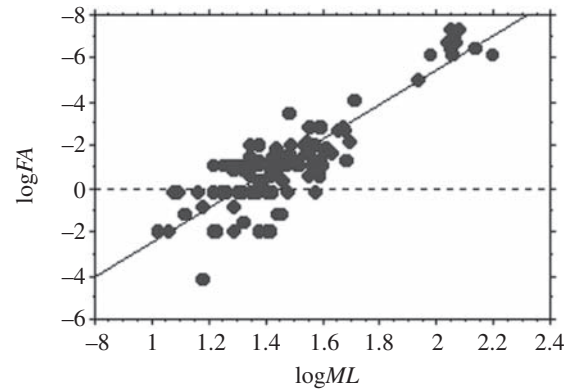

Fig. 6. Log diameter of funnel aperture (*FA*) *versus* log dorsal mantle length (*ML*). y=–1.053+0.803x; r^2=0.758, *F*-test P<0.0001. Because the coefficient of the log*ML* in the equation is <1, there is negative allometry of the funnel aperture with development; it is largest relative to the *ML* in the smallest squids.

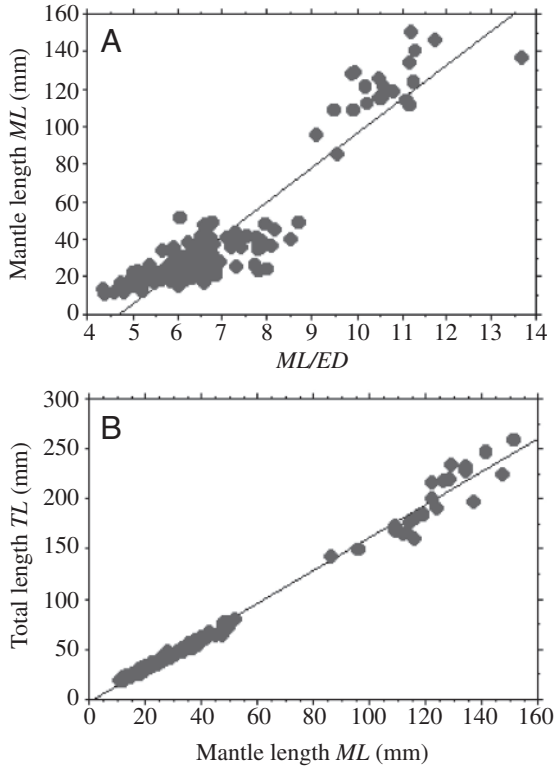

Fig. 7. *Loligo opalescens.* (A) Linear regression of dorsal mantle length (*ML*) from a ratio of *ML* upon eye diameter (*ED*). y=–85.285+18.205x; r^2=0.857, P<0.001, N=157. (B) Total length, the sum of the dorsal mantle length, the head length and the fourth arm length, can be estimated from the mantle length by the linear regression y=–4.0244+1.6227x; r^2=0.978, P<0.001, N=157.

Discussion

Juvenile market squid are undoubtedly difficult to capture with midwater trawls, but it is also likely that they do not often venture into midwater off the continental shelf. All of the video in which the juvenile *Loligo opalescens* occurred was over the Monterey shelf in 5–50 m of water at night. Further, all of the juvenile squid that I captured from March 1998 to February 2002 in Santa Monica Bay were also on the continental shelf, during the day, within 3 miles of the shore. I captured far more juvenile squid with much smaller nets over the continental shelf than did Mais (1974) using large nets in deeper waters. Therefore I speculate that juvenile *Loligo* of 15–55 mm *ML* remain over the continental shelf until they attain a size such that they can perform the vertical migrations to 500 m by day observed in the adults (Hunt, 1996). They may remain near the bottom by day (where they were captured by our small benthic otter trawl) and migrate up into the water column at night (where they were most often videotaped).

The behaviors of juvenile squid were similar to those observed in the adults (Hunt et al., 2000). All but three of the 13 adult chromatic components and two new ones were exhibited, the differences possibly due to incomplete development of the neuro-muscular controlled chromatophores

(Messenger, 2001). Juveniles displayed all of the seven adult postural components. It is important to note that Hunt et al. (2000) did not make observations of spawning adults and therefore the adult repertoire will contain more postures and chromatic displays than listed, for example spawning *Loligo opalescens* males usually display an all-white mantle and head, and bright red arms when clasping the female's mantle.

The initiation of acceleration from hovering is a movement that is similar to the fast starts (both C-starts and J-starts) performed by fish (Webb, 1994) to overcome inertial forces of starting movement from rest. Fish perform these motions within 50–200 ms, and squid make these starts within 130 ms. The C-start and J-start of fish are confined to the first two tail beats of the animal, while in squid there is only one flap of the arms and then the jetting takes over.

Bartol et al. (2001) proposed that small squids (<30 mm *ML*) could make use of a series of toroids to increase efficiency of jet propulsion. The vorticity of one ring draws in the water of the next ring, enhancing propulsion. I never observed more than one ring formed. I observed toroid formation only during the most extreme cases of deceleration, when squid stopped and changed directions in the midst of high velocities. Inking squid, observed to make directional changes while swimming at lower velocities, did not produce toroids.

O'Dor (1982) determined that adult squid can swim at a critical speed of 0.36 m s^{-1} and had a maximum speed of 1.01 m s^{-1} in a flow tunnel. However, a respirometer is a highly microturbulent environment, leading O'Dor to speculate that squid could reach speeds of 1.67 m s^{-1} in the laminar flow regime of the open ocean. I estimated that the greatest speed attained by any squid was 1.60 m s^{-1} for a *Loligo opalescens* of *ML*=116 mm. O'Dor (1988a) found that *Loligo opalescens* uses its fins in swimming only at speeds below 0.20 m s^{-1}. The average speed of our 26 analyzed sequences was 0.21 m s^{-1}, and the squid were using their fins intermittently. Perhaps squid were most often observed at this velocity because it is more metabolically efficient to swim with the use of a jet in conjunction with fins. Above this speed, thrust must come entirely from their jets.

The 0.20 m s^{-1} mean cruising speed for squids ranging from 12 to 116 mm *ML* has important implications for the migration capabilities and ecology of *Loligo opalescens*. The average speed was 5.15 *ML* s^{-1}, more than twice the previous estimate of optimum efficiency of 2 *ML* s^{-1} (O'Dor, 1988b), and thus estimates of possible migration capabilities of 600 km (assuming 12 h of travel/day) may be rather conservative. Reichow and Smith (2001) found that as few as 20 squid migrating from Monterey Bay to southern California could account for the genetic homogeneity encountered in microsatellite DNA samples of this species. *Loligo opalescens* may live their entire lives within a few miles of the shores near which they were hatched. But given the swimming capabilities, lifespan and a push from the California Current (equatorward) or Davidson Current (poleward), it is possible that the populations of market squid in northern and southern California commonly exchange genetic information *via* migration.

Based upon the linear regression of *ML*=–14.7–0.627×Age (Butler et al., 1999), the squid in our video sequences were 88±42 days old (mean ± S.D., range 39–209 days, *N*=26).

Paralarval and juvenile squid swimming is dominated by 'jet-and-sink' swimming, where the fins merely stabilize. As squid develop, their motions transform to 'climb-and-glide' swimming, in which fins contribute substantially to the generation of thrust (O'Dor, 1988a). The growth of the organism's diameter and thrust-producing capabilities facilitate the transition from the viscosity-dominated environment of the hatchling to the inertial realm of the adult. The effect of water viscosity on squid of juvenile and adult size is readily apparent when comparing the swimming videos [movie 4, juvenile_swimming and 5, adult_swimming; available in supplementary material; video footage ©1998, ©2000 MBARI]. Future use of allometric measurements for scale to examine Reynolds numbers may prove useful in the explanation for this behavioral transition. It will be important to use different diameters for mantle width when the squid is performing inhalation and exhalation. Since the velocity fluctuates largely with mantle contractions, *Re* numbers will similarly fluctuate widely as the squids swim with the unsteady aspects of teuthid jet propulsion (Anderson and Demont, 2000; Hunt et al., 2000; Daniel, 1984).

Previously, the analysis of jet and sink behavior has been impossible in the laboratory because the small horizontal space of the respirometer's working section did not allow the analysis of 'hovering which usually involves nearly vertical oscillations of several centimeters' (O'Dor, 1982). Analysis of ROV video sequences allows observations of squid swimming in all directions, both against currents and in still water. Webber et al. (2000) reported the intra-mantle pressure and 100 m vertical migration of a telemetered *Loligo forbesi*. The squid sustained higher pressures during the climb than during the descent, indicating the increased amount of energy required to move up in the water column. Even on a small scale, it is evident from the size of the mantle contraction that the energetic requirements of moving up in the water column for a negatively buoyant squid can be substantial. Squids may migrate vertically, perhaps to follow prey, to avoid diurnal visual predators (Hardy and Bainbridge, 1954) or to lower their metabolic rate (O'Dor et al., 1986).

Boletsky (1987) stated that the small fins of hatchlings provide little or no dynamic lift, but may counteract the tilting effect of the funnel jet expelled on the ventral side. Hoar et al. (1994) concur, using a ratio of fin length to dorsal mantle length that increases from 0.17–0.47 in *Loligo opalescens* with ontogeny. Considering that viscous forces dominate the flow regime of hatchlings, the researchers speculate that the fins serve to provide stabilization only.

Perhaps there is an adaptive reason for the lobate fin shape and small fin size of the hatchlings. The fins may be so small because that is all that is required to provide lift for a smaller animal. More likely, the apparent lack of use of the fins in hatchlings may not be a factor of their size. It may be due to a lack of neurological control. In the first 2 months of life,

squid paralarvae have been shown to develop hunting behaviors (Chen et al., 1996), change in mantle musculature composition (Preuss et al., 1997; Thompson and Kier, 2001a), and develop different jet escape responses (Preuss and Gilly, 2000; Thompson and Kier, 2001b). Even more likely, it is a matter of scale. At 3 mm *ML* a hatchling lives in a highly viscous environment, and fin flapping at this Reynolds number would require much greater rigidity, enervation and musculature than is present for such a negatively buoyant organism; thus jetting is more efficient. O'Dor and Webber (1986) suggest that jet propulsion is more efficient than undulation at these sizes.

The funnel aperture diameter has a negative allometric growth factor of 0.803. Squid attain their thrust from a small cross-sectional area of water being rapidly expelled (Wells and O'Dor, 1991). The increasingly smaller aperture of the developing squid is probably a function of the developing mantle musculature (Preuss et al., 1997; Thompson and Kier, 2001a). The jet aperture gets proportionately smaller as the squid develops greater mantle-contracting abilities, and can force larger volumes of water faster through a tighter hole, with higher pressures.

Underwater video and photography is becoming increasingly commonplace as technology becomes more affordable. This video was captured by the Monterey Bay Aquarium Research Institute ROV *Ventana*, a multi-million dollar vehicle that can log the focus telemetry and zoom telemetry of the camera lens for all images (Davis and Pilskaln, 1993). However, the development of allometry measurements to provide scale for photographic images could be used with a hand-held digital camera. This technique can be used to assess the health of underwater environments. The regression reported herein for squid could easily be applied to fish or other animals. Currently there is a worldwide program to assess the health of coral reefs, 'Reef Check' (Hodgson, 2001). The use of this allometry scale can add the parameter of fish age to the already collected data of fish counts. Reefs that are covered with juvenile fish due to over-fishing of the adults could give the false impression of a healthy reef ecosystem, based upon fish counts alone. Still photography of fish schools could provide images of the profiles of large numbers of fish. A comparison of eye diameter to total length of the fish in the photograph could be used in conjunction with an allometric series for a given species to estimate the length, sex (if there is dimorphism) and ages of the fish, and from this the biomass. This could be an index of the anthropogenic forcing placed upon the reef. This technique could be applied to any species with conspicuous features and allometric growth. Currently, many species of salmonids are monitored with photographs taken as fish are forced to swim through traps and ladders. In addition to checking for hatchery fin clips, allometry regression could be used to estimate the size and age of these fish and to distinguish between hatchery-reared and wild-stock salmon.

I am grateful to William H. Hamner, Bruce Robison, Dean Lauritzen, Richard Ambrose, Peggy Fong, Dick Zimmer, Beverly Macewicz and Dan Davis for editorial comments. Alex Rappaport assisted with allometry measurements; Gwen Noda and Dennis Weyrauch collected the trawled squid. The R/V *SeaWorld UCLA* was a gift from Milton and Peggie Shedd. This research was financed with grants from Coastal Environmental Quality Initiative Program no. 783828-KH-19900, and supported in part by the David and Lucile Packard Foundation through Monterey Bay Aquarium Research Institute.

References

Anderson, E. J. and Demont, M. E. (2000). The mechanics of locomotion in the squid *Loligo pealei*: locomotory function and unsteady hydrodynamics of the jet and intramantle pressure. *J. Exp. Biol.* **203**, 2851-2863.

Bartol, I. K., Patterson, M. R. and Mann, R. (2001). Swimming mechanics and behavior of the shallow-water brief squid *Lolliguncula brevis*. *J. Exp. Biol.* **204**, 3655-3682.

Boletsky, S. V. (1987). Juvenile behavior. In *Cephalopod Life Cycles*, Vol. 2. *Comparative Reviews* (ed. P. R. Boyle), pp. 45-60. London: Academic Press.

Butler, J., Fuller, D. and Yaremko, M. (1999). Age and growth of market squid (*Loligo opalescens*) off California during 1998. *Calif. Coop. Oceanic Fish Invest. Rep.* **40**, 191-195.

Chen, D. S., Van Dykhuizen, G., Hodge, J. and Gilly, W. F. (1996). Ontogeny of copepod predation in juvenile squid (*Loligo opalescens*). *Biol. Bull.* **190**, 69-81.

Daniel, T. L. (1984). Unsteady aspects of locomotion. *Am. Zool.* **24**, 121-134.

Davis, D. L. and Pilskaln, C. H. (1993). Measurements with underwater video: camera field width calibration and structured lighting. *MTS J.* **26**, 13-19.

Denny, M. W. (1993). *Air and Water the Biology and Physics of Life's Media.* Princeton, NJ: Princeton University Press.

Hardy, A. C. and Bainbridge, R. (1954). Experimental observations on the vertical migration of plankton animals. *J. Mar. Biol. Assn. (UK)* **33**, 409-448.

Hatfield, E. M. C., Hanlon, R. T., Forsythe, J. W. and Grist, E. P. M. (2001). Laboratory testing of a growth hypothesis for juvenile squid *Loligo pealeii* (Cephalopoda: Loliginidae) *Can. J. Fish. Aquat. Sci*. **58**, 845-857.

Hoar, J. A., Sim, E., Webber, D. M. and O'Dor, R. K. (1994). The role of fins in the competition between squid and fish. In *Mechanics and Physiology of Animal Swimming* (ed. L. Maddock, Q. Bone and J. M. V. Rayner), pp. 27-43. New York: Cambridge University Press.

Hodgson, G. (2001). Reef check: The first step in community-based management. *Bull. Mar. Sci.* **69**, 861-868.

Hunt, J. C. (1996). The behavior and ecology of midwater cephalopods from Monterey Bay: submersible and laboratory investigations. PhD thesis, University of California, Los Angeles, USA.

Hunt, J. C. and Seibel, B. A. (2000). Life history of *Gonatus onyx* (Cephalopoda: Teuthoidea): ontogenetic changes in habitat, behavior and physiology. *Mar. Biol.* **136**, 543-552.

Hunt, J. C., Zeidberg, L. D., Hamner, W. M. and Robison, B. H. (2000). The behavior of *Loligo opalescens* (Mollusca: Cephalopoda) as observed by a remotely operated vehicle (ROV) *J. Mar. Biol. Assn. (UK)* **80**, 873-883.

Jackson, G. D. and Domeier, M. L. (2003). The effects of an extraordinary El Niño/La Niña event on the size and growth of the squid *Loligo opalescens* off Southern California. *Mar. Biol.* **142**, 925-935.

Mais, K. F. (1974). Pelagic fish surveys in the California current. *Fish Bull.* **162**, 1-79.

Messenger, J. B. (2001). Cephalopod chromatophores: neurobiology and natural history. *Biol. Rev.* **76**, 473-528.

Moltschaniwskyj, N. A. (1995). Changes in shape associated with growth in the Loliginid squid *Photololigo* sp.: a morphometric approach. *Can. J. Zool.* **73**, 1335-1343.

O'Dor, R. K. (1982). Respiratory metabolism and swimming performance of the squid, *Loligo opalescens*. *Can. J. Fish. Aquat. Sci*. **39**, 580-587.

O'Dor, R. K. (1988a). Forces acting on swimming squid. *J. Exp. Biol.* **137**, 421-442.

O'Dor, R. K. (1988b). The energetic limits on squid distributions. *Malacologia* **29**, 113-119.

O'Dor, R. K. and Hoar, J. A. (2000). Does geometry limit squid growth? *ICES J. Mar. Sci.* **57**, 8-14.

O'Dor, R. K. and Webber, D. M. (1986). The constraints on cephalopods: why squid aren't fish. *Can. J. Zool.* **64**, 1591-1605.

O'Dor, R. K., Foy, E. A. and Helm, P. L. (1986). Locomotion and energetics of hatchling squid, *Illex illecebrosus*. *Amer. Malacol. Bull.* **4**, 55-60.

Okutani, T. (1987). Juvenile morphology. In *Cephalopod Life Cycles,* Vol. 2. *Comparative Reviews* (ed. P. R. Boyle), pp. 33-44. London: Academic Press.

Packard, A. (1969). Jet propulsion and the giant fibre response of *Loligo*. *Nature* **221**, 875-877.

Preuss, T., Lebaric, Z. N. and Gilly, W. F. (1997). Post hatchling development of the circular mantle muscles in the squid *Loligo opalescens*. *Biol. Bull.* **192**, 375-387.

Preuss, T. and Gilly, W. F. (2000). Role of prey-capture experience in the development of the escape response in the squid *Loligo opalescens*: a physiological correlate in an identified neuron. *J. Exp. Biol.* **203**, 559-565.

Reichow, D. and Smith, M. (2001). Microsatellites reveal high levels of gene flow among populations of the California squid *Loligo opalescens*. *Mol. Ecol.* **10**, 1101-1109.

Rodhouse, P. G. and Piatkowski, U. (1995). Fine-scale distribution of juvenile cephalopods in the Scotia Sea and adaptive allometry of the brachial crown. *Mar. Biol.* **124**, 111-117.

Thompson, J. T. and Kier, W. M. (2001a). Ontogenetic changes in fibrous connective tissue organization in the oval squid, *Sepioteuthis lessoniana* Lesson, 1830. *Biol. Bull.* **201**, 136-153.

Thompson, J. T. and Kier, W. M. (2001b). Ontogenetic changes in mantle kinematics during escape-jet locomotion in the oval squid, *Sepioteuthis lessoniana* Lesson, 1830. *Biol. Bull.* **201**, 154-166.

Thompson, J. T. and Kier, W. M. (2002). Ontogeny of squid mantle function: changes in the mechanics of escape-jet locomotion in the oval squid, *Sepioteuthis lessoniana* Lesson, 1830. *Biol. Bull.* **203**, 14-26.

Vidal, E. A. G. (1994). Relative growth of paralarvae and juveniles of *Illex argentinus* (Castellanos, 1960) in southern Brazil. *Ant. Sci.* **6**, 275-282.

Webb, P. W. (1994). Exercise Performance of Fish. In *Advances in Veterinary Science and Comparative Medicine,* Vol. 38B. *Comparative Vertebrate Exercise Physiology: Phyletic Adaptations* (ed. J. J. Jones), pp. 1-49. San Diego: Academic Press.

Webber, D. M., Aitken, J. P. and O'Dor, R. K. (2000). Costs of vertic dynamics of cephalopods and fish. *Physiol. Biochem. Zool.* **76**, 651-662.

Wells, M. J. and O'Dor, R. K. (1991). Jet propulsion and the evolution of the cephalopods. *Bull. Mar. Sci.* **49**, 419-432.

Wormuth, J. H. and Roper, C. F. E. (1983). Quantitative sampling of oceanic cephalopods by nets: problems and recommendations. *Biol. Oceanog.* **2**, 357-377.

Yang, W. T., Hixon, R. F., Turk, P. E., Krejci, M. E., Hulet, W. H. and Hanlon, R. T. (1986). Growth, behavior, and sexual reproduction of the market squid, *Loligo opalescens*, cultured through the life cycle. *Fish. Bull.* **84**, 771-798.

Zeidberg, L. D. and Hamner, W. M. (2002). Distribution of squid paralarvae, *Loligo opalescens* (Cephalopoda: Myopsida), in the Southern California Bight in the three years following the 1997 El Niño. *Mar. Biol.* **141**, 111-122.

Zuev, G. V. (1964). On the ability of cephalopod larvae to active movement. *Zool. Zh.* **43**, 1440-1445.